

## Liver Allo- and Xenotransplantation

T.E. Starzl

**P**ERHAPS I should start by saying that I owe my share of this prize to the people with whom I have worked, and especially to the patients who allowed us to treat them.

I came into transplantation through the back door of physiology, during metabolic investigations of portal physiology in the mid-1950s. By 1959, we had clarified the surgical secrets of liver transplantation.<sup>1</sup> These were (1) the hepatotrophic (liver-supporting) superiority of portal venous blood that was required for revascularization; (2) the need for core cooling of the liver graft as is practiced today as the first step in the preservation of all organs; and (3) the use of a venovenous bypass during the anhepatic period. Unknown to us at first, Francis D. Moore's team in Boston had begun similar liver transplant research at the same time and had come to many of the same conclusions.<sup>2</sup> Our work on liver transplantation was finished by the end of 1959, as was a second project in dogs on multivisceral transplantation,<sup>3</sup> which 25 years later was performed successfully in humans and was the basis for several variations, such as cluster and liver-intestine transplantations.<sup>4</sup>

This activity in 1958 through early 1960 was in a therapeutic vacuum because there was no such thing as practical immunosuppression. The classical paper on 6-mercaptopurine by Schwartz and Dameshek<sup>5</sup> in nontransplant models and its testing in skin and kidney transplant models<sup>6-9</sup> were published toward the end of this period. Realizing by now that the road to the liver would have to be through the simpler kidney transplant model, I moved from Northwestern University in Chicago to the University of Colorado in 1961 in order to begin a clinical kidney program.

The Colorado kidney program was based on the simple laboratory discovery that canine kidney rejection under azathioprine could be reversed with prednisone in 88% of dogs, a rate that proved to be the same in humans.<sup>10</sup> Neither azathioprine nor adrenal cortical steroids in dogs or humans allowed kidney transplantation to be performed at a practical level,<sup>11</sup> but the drug combination became the basis for a clinical specialty and led to two further initiatives, both of which failed. The first was liver transplantation, which was technically successful in two of the first three cases,<sup>12</sup> but resulted in death in all patients. The second initiative was baboon-to-human kidney xenotransplantation,<sup>13</sup> following Reemtsma's pathfinding trials with chimpanzee donors.<sup>14</sup> The baboon kidneys were rejected

after 6 to 60 days, foreclosing further baboon trials for more than two decades.

The liver program was itself closed for 4 more years of intensive further laboratory investigation. Then, in July 1967, the first long-surviving liver recipients were produced<sup>15</sup> in a collaboration with Carl Groth of Stockholm, who was a key team member during this effort, which by now had consumed almost 10 years. Acceptance of the procedure was slow over the next dozen years because of its high mortality. Clinical liver transplantation became a curiosity, being performed by a few stubborn eccentrics on both sides of the Atlantic. Roy Calne's introduction of cyclosporine<sup>16</sup> allowed a tripling of survival about a decade ago and brought liver transplantation to center stage.<sup>17</sup>

Recently the liver has been the lead organ in understanding the immunologic and metabolic terms of successful xenotransplantation. Following hamster-rat xenotransplantation, the interstitial leukocytes that I discussed in my Presidential Address migrate ubiquitously (Fig 1), whereas the hepatocytes retain their donor metabolic specificity and recreate their own chemical environment. For example, the serum proteins of the rat become those of the hamster within 1 week after xenotransplantation of a hamster liver (Fig 2). The hamster liver also brings its own coagulation profile. For one thing, the rat, which does not produce a supply of the natural anticoagulant, C protein, acquires the C protein and is "hamsterized" in other ways within days of receipt of a hamster liver. Yet, neither bleeding nor clotting were encountered. This observation provided the final assurance leading to a decision to proceed with a clinical trial.

The same transition of serum proteins to that of the donor occurred in our human recipient of a baboon liver. The metabolic consequences are so wide-ranging that years will be required to study them completely. An example is serum uric acid, which is near zero in the normal baboon whose liver has well-developed enzyme systems for conversion of this metabolite. After xenotransplantation of a baboon liver, there was virtual disappearance of uric acid from the patient's serum.

---

From the Department of Surgery, University of Pittsburgh, Pennsylvania.

Address reprint requests to Dr Thomas E. Starzl, Department of Surgery, 3601 Fifth Ave, Falk Clinic, Pittsburgh, PA 15213.

© 1993 by Appleton & Lange  
0041-1345/93/\$3.00/+0

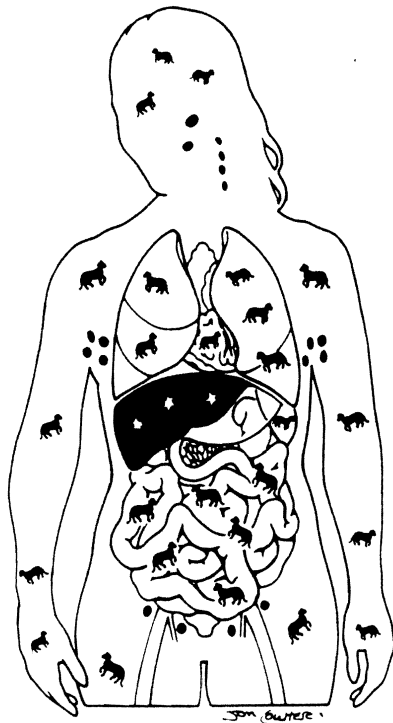


Fig 1. Nature of chimerism after xenotransplantation.

The baboon-to-human liver xenotransplantation was performed in Pittsburgh on June 28, 1992 (by Drs Andreas Tzakis, John Fung, and Satoru Todo). The recipient is a 35-year-old man with end-stage chronic active hepatitis

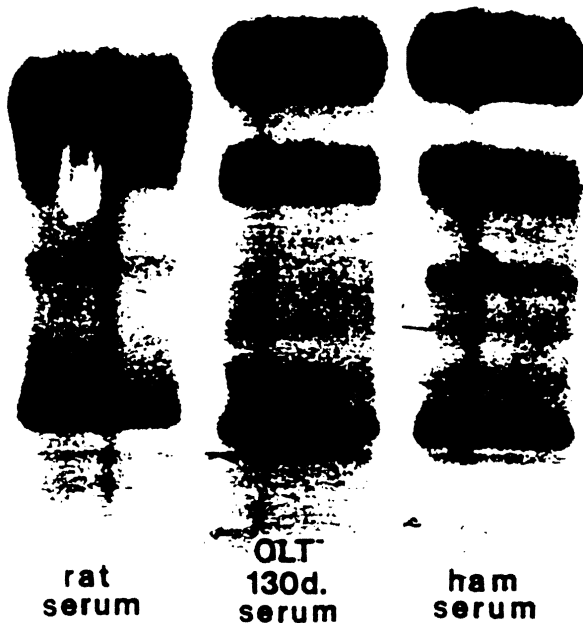


Fig 2. Serum protein electrophoresis. Albumin, dark upper band (SPE kit, Beckman Instruments, Brea, CA).

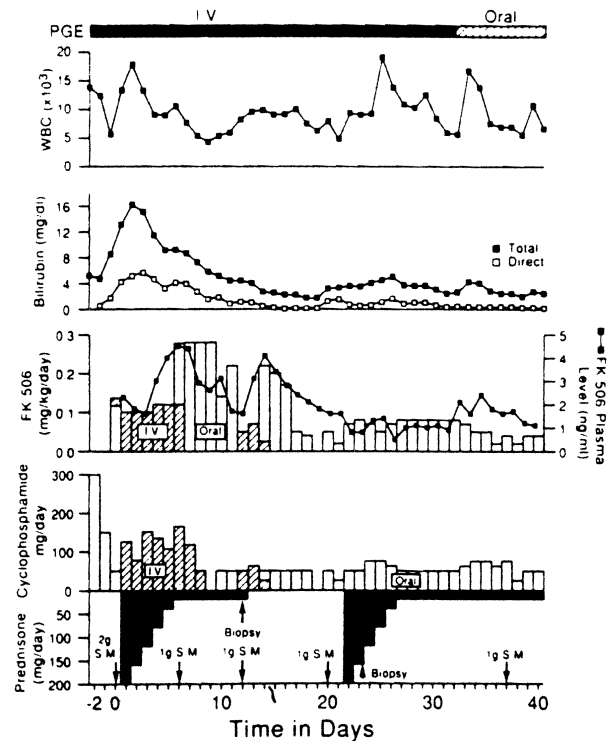


Fig 3. Course of patient after receipt of baboon liver. PGE, prostaglandin E.

caused by the B virus that is thought incapable of infecting the baboon liver. Baseline immunosuppression was with conventional doses of FK 506, prednisone, and prostaglandin E, essentially the same formula as our preferred therapy after hepatic allotransplantation (Fig 3). Cyclophosphamide in nontoxic doses was added, based on the investigations by Murase et al<sup>18</sup> showing amelioration of the xenograft humoral rejection with this drug. Serious rejection has never been encountered. The patient's bilirubin is normal.

A postperfusion biopsy of the baboon liver from a 53-pound donor showed neutrophils in the sinusoids but no clinical evidence of hyperacute rejection. Twelve days later, the liver looked grossly normal and on biopsy showed a minor periportal cellular infiltrate.

Although biopsies specimens taken at 12 and 24 days showed only minimal cellular rejection and were almost normal except for mild cholestasis, immunofluorescence at 12 days showed extensive deposits of immunoglobulin M (IgM), which at 24 days had largely disappeared. The IgG followed the same pattern. Meanwhile, by day 24 the liver had regenerated to the appropriate size for the 170-pound recipient.

The patient is well 53 days posttransplantation. We know that he has started the transformation to a state of mixed chimerism that we believe is integral to xenograft as well as allograft acceptance. At 35 days, blood chimerism was identified with baboon chorionic gonadotropin B sub-

unit genes. Normal human blood has no baboon marker, but this patient has had a striking baboon band, estimated from dilution studies to be one baboon cell/1,000 human cells.

It is too early to be sure, but it looks now as if the immune reaction to the baboon liver can be controlled without toxic immunosuppression. There do not appear to be serious metabolic incompatibilities with the baboon-human species combination. There is no evidence yet of reinfection of the liver with B virus. If this resistance continues, there are implications for other viral infections with which baboons cells also cannot be infected, including human immunodeficiency virus.

Should this effort remain successful, it will be the vindication of yet another vision of Peter Medawar, who had a long interest in xenotransplantation. Almost 400 years ago to the day, Galileo arrived to his faculty position at the University of Padova. Armed with a telescope, he began the inquiries that defined the mysterious universe and led to a walk on the moon. Our Galileo was Peter Medawar, whose first inquiries into the biologic meaning of rejection and the universe of transplantation took place 12½% of the time back to those medieval days. For me, there could be no greater honor than to receive a prize named Medawar.

#### REFERENCES

1. Starzl TE, Kaupp HA Jr, Brock DR, et al: Surg Gynecol Obstet 111:733, 1960
2. Moore FD, Wheeler HB, Demissianos HV, et al: Ann Surg 152:374, 1960
3. Starzl TE, Kaupp HA Jr: Surg Forum 11:28, 1960
4. Starzl TE, Todo S, Tzakis A, et al: Surg Gynecol Obstet 172:335, 1991
5. Schwartz R, Dameshek W: Nature 183:1682, 1959
6. Meeker W, Condie R, Weiner D, et al: Proc Soc Exp Biol Med 102:459, 1959
7. Schwartz R, Dameshek W: J Clin Invest 39:952, 1960
8. Calne RY: Lancet 1:417, 1960
9. Zukoski CF, Lee HM, Hume DM: Surg Forum 11:470, 1960
10. Starzl TE, Marchioro TL, Waddell WR: Surg Gynecol Obstet 117:385, 1963
11. Murray JE, Merrill JP, Harrison JH, et al: N Engl J Med 268:1315, 1963
12. Starzl TE, Marchioro TL, Von Kaulla KN, et al: Surg Gynecol Obstet 117:659, 1963
13. Starzl TE, Marchioro TL, Peters GN, et al: Transplantation 2:752, 1964
14. Reemtsma K, McCracken BH, Schlegel JU, et al: Ann Surg 160:2:752, 1964
15. Starzl TE, Groth CG, Brettschneider L, et al: Ann Surg 168:392, 1968
16. Calne RY, Rolles K, White DJG, et al: Lancet 2:1033, 1979
17. Starzl TE, Klintmalm GBG, Porter KA, et al: N Engl J Med 305:266, 1981
18. Murase N, Starzl TE, Demetris AJ, et al: Transplantation (in press)